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A microscopic study of cross-sections of woody dicotyledonous stems with reference to their use in courses in elementary botany

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A MICROSCOPIC STUDY OF CROSS-SECTIONS OF WOODY DICOTYLEDONOUS STEMS WITH REFERENCE TO THEIR USE IN COURSES IN ELEMENTARY BOTANY

By

Nancy Jane Toms

June 1, 1936

To

Dr. Ernest Elwood Stanford

for his invaluable assistance

and friendship throughout my

college years.

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CHAPTER I

INTRODUCTION

Statement of the Problem

The problem of this study was to discover a woody dicotyledonous stem which could be used as the principal example of stem structure in a course in elementary botany.

Considerations which Govern the Study of the Woody Stem

Significance of the stem

The stem is one of the primary organs of the plant body. It connects the roots, which are the absorbing organs and the leaves, which are the manufacturing organs. It provides an avenue of transportation between these. It also serves to support the leaves and flowers in situations favorable for their functions. The stem is a conspicuous feature of most plants, particularly of woody species where it constitutes the great bulk of their bodies.

The stem as a subject of study

The woody stem of the dicotyledon is a complex organ composed of many kinds of cells with much variety in form, function and arrangement. Its cells represent the principal cell types of the plant as a whole. The cells of the plant body are grouped, according to similarity in origin, form and function, into tissues. In the woody stem the tissues are arranged in a definite manner and are continuous throughout the other plant organs. Therefore, the stem with its varied and representative cellular structures and its organization

into definite tissues may serve as a principal illustration of cellular structure and organization in plants. Its study should, of course, be preceded by some preliminary study of the plant cell, and by some attention to the simpler gymnosperm stem.

Relation of the study of the woody stem to that of more advanced stem-types

Study of the woody stem serves as a basis for the understanding of organization of other types of stems.

The three principal types of stem structure are the woody stem, the herbaceous stem and the monocotyledonous stem. The perennial woody stem is the most ancient stem-type. In such a stem the vascular tissue forms a continuous and rather wide ring, which is added to indefinitely by cambial activity. The other stem types are believed to have descended from woody types through evolutionary changes and simplifications.

In the herbaceous type, where the stems are much softer and shorter lived, the vascular tissue has been reduced and broken up. However, the majority of forms possess cylinders of vascular tissue that are complete except for leaf and branch gaps.[1] The herbaceous stem is essentially similar to, though smaller and simpler than, the woody stem and may well be studied after that.

The monocotyledonous type of stem, with the vascular bundles widely distributed through the stele, represents the

[1] Eames and MacDaniels, An Introduction to Plant Anatomy, 245.

extreme condition in reduction of vascular tissue and cambial activity from the woody type, and may properly be studied after the woody and herbaceous types have received attention.

The stem in its relation to the cellular structure of the plant in general

Study of the woody stem serves as a basis for an understanding of cellular arrangements and relationships in other parts of plants. The root, stem, and leaves of the plant body are not independent organs, but are interdependent parts of the whole organism. The three main tissue systems, the epidermis, the cortex, and the fibro-vascular system are continuous throughout.

The epidermis in all plant organs consists of a single integumentary layer of cells. The cortex lies directly beneath the epidermis and, although it varies in form and function in the different plant organs, it is the fundamental tissue. The fibro-vascular system is continuous throughout the plant and is in a sense concentrated in the stem. Therefore, the study of this organ is a desirable preliminary to that of the tissue differentiations in other plant parts.

Structure and function in the stem.

Study of the woody stem affords an opportunity for emphasis on relationships between structure and function. The primary functions of the stem are the support of the leaves and flower parts, the conduction of water and minerals from the soil, and the conduction of foods from the manufacturing

organs to the points where they are needed in growth or where they will be stored for future use. In addition, the stem frequently acts as an organ of food storage and food manufacture. These varied activities are performed by specialized cells within tissue regions of the stem, and in the study of this organ, the functions of these microscopically visible cells and tissues may profitably be stressed and their visible characteristics correlated with some simple demonstrations of a physiological nature.

Stem study as a basis for the comprehension of mechanical characteristics and values of wood

In the stem are several types of tissue whose functions are mechanical in nature. The sclerenchyma cells and fibrous tissues have thick lignified walls. Such tissues support the whole plant body. Those commercial timbers called hardwoods, are made up chiefly of lignified, or wood, fibers. Most of the other cell types of the central portion of the stem are lignified also. The heavy cell walls and the narrow cavities, which contain air, impart to the wood the characteristics of strength, rigidity and flexibility which make it valuable as a structural material.

Qualities Desirable in a Woody Stem for Class Use

The woody stem to be used for class study should be of a familiar species or one to which the student may readily be introduced. Living specimens should be available for study in conjunction with the prepared sections. Interest

may be added also if the trees are sufficiently near at hand so that students may become familiar with them.

The tissues of a suitable woody stem should be of fairly "typical" kinds, that is, kinds that are likely to be found in most plant stems. They should be easily recognizable and well differentiated from each other, and arranged in a manner which may be readily represented by diagrams.

It is desirable to have a woody stem which can be embedded and cut by standard processes. Both wood and bark tissues should hold together when cut in fairly thin sections. The stem should not be so tough and hard that it cannot be cut without injury to the microtome knife.

Stem specimens should be of size suitable to show more than one growth ring, for the study of both spring and summer tissues.

CHAPTER II

TECHNICAL PROCEDURE

Preparation and Fixation of Material

Two year old twigs were selected from trees growing on the campus of the College of the Pacific and cut into pieces from one-quarter to three-eighths of an inch in length.

Some of the stem specimens were killed and fixed in formalin acetic alcohol made by using:

Glacial acetic acid.....	5.00 c.c.
Commercial formalin.....	5.00 c.c.
70 per cent alcohol.....	90.00 c.c.[1]

The remainder were put in a mixture of:

Distilled water.....	100.00 c.c.
Chromic acid.....	.75 g.
Glacial acetic acid.....	1.00 c.c.
Urea.....	.50 g.[2]

The materials were left in these solutions for several days to insure thorough infiltration of the reagents.

Excess of the alcohol fixative was washed out with frequent changes of 70 per cent alcohol; excess of the chrome-acetic acid mixture with a gentle stream of running water for about 24 hours.

The materials while still in water were placed in a sealed vacuum chamber and a vacuum pump attached in order to

[1] Chamberlain, Methods in Plant Histology, 21.

[2] H. P. Brown, Personal Letter, Sept. 25, 1935.

remove the air from the tissues so that they might be thoroughly impregnated with parlodion. When the materials sank, they were removed.

For softening, the harder woods were placed for a period of two weeks in paraffin covered bottles containing full strength hydrofluoric acid. Softer woods were placed in 25 to 50 per cent aqueous solutions of the acid, and left for a week or ten days.

After treatment with the hydrofluoric acid, the stems were washed thoroughly in water for 25 to 48 hours to remove all traces of the acid.

The softer materials were dehydrated in the following grades of ethyl alcohol: 15, 30, 50, 70, 85, 95, and absolute. Absolute alcohol was changed once. The time in each grade was from 12 to 24 hours.

The harder woods were dehydrated by the butyl alcohol method,[1] in order to avoid the hardening effects of ethyl alcohol. Zirkle's[2] procedure for specimens containing cambium and phloem as well as xylem was used. The following numbers indicate the percentages:

Water.....	95	89	82	70	50	30	15	5	0	0	0
Ethyl alcohol.....	5	11	18	30	40	50	50	40	25	0	0
Butyl alcohol.....	0	0	0	0	10	20	35	55	75	100	100

1 P. H. Wetmore, "The Use of Celloidin in Botanical Technique", reprinted from Stain Technology, 7, (1932), No. 2, 52.

2 Conway Zirkle, "The Use of n-Butyl Alcohol in Dehydrating Woody Tissue for Paraffin Embedding", Science, 71, (1930), 103.

The materials were left one hour in each medium except for the final change in 100 per cent butyl alcohol, in which they were left over night.

Embedding and Hardening

In order to facilitate embedding, the materials were first immersed in ether alcohol (a mixture of equal parts of absolute alcohol and anhydrous ether) for 24 hours. Parlodion was dissolved in ether alcohol; concentrations of 2, 4, 6, 8, 10, 12 per cent being used.

The materials were placed in small bottles with wide mouths and caps that could be screwed down tightly. The bottles were filled about one-half full of 2 per cent parlodion, sealed and placed in a constant temperature oven at about 50°C. They were left over night or longer, then removed and cooled, after which the 2 per cent parlodion was poured off and replaced by 4 per cent. This procedure was continued through all of the grades of parlodion mentioned. It was found that after a day or two in the oven the 12 per cent parlodion became thick enough for hardening.

The pieces of stems were removed from the bottles, still surrounded by parlodion, and dipped in chloroform for a few minutes; then left over night in chloroform. The blocks were then stored in a preserving and softening solution of equal parts by volume of glycerine and 95 per cent ethyl alcohol until ready to be trimmed and cut.

White pine blocks 1 inch long and $\frac{1}{2}$ inch by $\frac{1}{2}$ inch in cross section were used as object carriers. One end of the

block was dipped in ether-alcohol for a moment then in 6 per cent parlodion and allowed to dry. After the parlodion had set, the block of wood and the specimen block, which had previously been trimmed of excess parlodion, were both placed in 6 per cent parlodion for a few moments, then withdrawn and the specimen fixed on the wood block and allowed to set. They were placed in glycerine-alcohol until ready to use.

Staining and Mounting

A sliding microtome with a heavy knife was used for sectioning. The knife was adjusted at an oblique angle. The object and the knife were kept wet with 70 per cent alcohol. The sections cut were 25 to 40 microns in thickness and were removed from the knife blade with a camel's hair brush wet in 70 per cent alcohol and transferred to a watch glass of alcohol of the same percentage.

The sections of all the stem specimens, except one, were stained with Delafield's haematoxylin and eosin. The following schedule as given by Chamberlain[1] was used:

1. The sections were placed in watch glasses and covered with 70 per cent alcohol for 2 to 5 minutes.

2. They were then immersed in Delafield's haematoxylin for 5 to 30 minutes.

3. The sections were removed from the stain and washed in water for 5 minutes.

4. They were destained in acid alcohol (1 c.c. hydro-

[1] Chamberlain, Methods in Plant Histology, 127.

chloric acid and 100 c.c. of 70 per cent alcohol) until the stain was extracted from the parlodion, or until only a pink color remained.

5. The acid alcohol was washed out in 70 per cent alcohol until the purple color returned.

6. The sections were counter-stained in eosin (1 per cent solution in 70 per cent alcohol) for 2 to 5 minutes.

7. They were dehydrated in 95 per cent alcohol for 2 to 5 minutes.

8. Clearing was done in Eycleshmyer's clearing fluid for 1 or 2 minutes.

9. The sections were mounted in balsam on the slide, covered with a cover slip and put aside to dry.

As an experiment the specimen of Sycamore was stained in Haidenhain's iron-alum haematoxylin according to a schedule recommended by Herbert F. Marco[1] of the Wood Anatomy Laboratory of Yale University. The schedule was as follows:

1. The 70 per cent alcohol on the sections was replaced with 50 per cent alcohol, then 35 per cent, 25 per cent, 15 per cent, and distilled water.

2. The distilled water was drawn off and sections covered with a 4 per cent solution of ferric ammonium sulphate which acts as a mordant. Sections were left from 15 to 60 minutes.

3. The mordant was drawn off and sections washed in

[1] Herbert F. Marco, Circular Letter, 5.

four changes of distilled water. All excess mordant had to be removed to prevent precipitation with the reagent of the following step.

4. To the last wash water were added two or three drops of Haidenhain's haematoxylin. The reaction of the stain was watched under the microscope, and as soon as the middle lamella showed the stain, the solution was drawn off and the sections washed rapidly in 4 changes of distilled water.

5. The sections were dehydrated by washing successively in 15, 35, 50, and 70 per cent alcohols.

6. The alcohol was drained off and the sections counterstained in safranin for 30 to 60 minutes.

7. After staining the sections were washed in 70 per cent alcohol to remove excess stain. They were destained rapidly in acid alcohol and watched under the microscope.

8. The sections were dehydrated in several changes of 95 per cent alcohol and several changes of absolute alcohol to which a few drops of chloroform had been added to preserve the parlodion matrix.

9. The sections were cleared in xylol and mounted in balsam.

It was found advisable to place weights on the cover slips after mounting to flatten the sections so that they might be used for photographic purposes.

CHAPTER III

EXAMINATION OF STEMS

Tissues of the Woody Stem

The internal structure of the woody stem is typically composed of the following tissues, beginning at the outside: epidermis, cork, cortex, pericycle, phloem, cambium, xylem, vascular rays and pith. The epidermis, a single layer of cells, the cork, layers of suberized cells laid down by the cork cambium, and the cortex of parenchyma cells, make up the outer bark. The pericycle consisting of bast fibers and stone cells lends support and toughness to the stem. The phloem is made up largely of sieve tubes and companion cells which function in food conduction. The cambium is meristematic and provides for new growth in diameter of the stem. The xylem is made up of water conducting vessels, usually some tracheids, and wood fibers. Wood parenchyma may be more or less evident. Vascular rays, consisting of xylem and phloem portions appear in section as radial stripes, narrow in the xylem and broadening in the phloem. These function in storage and to some degree in conduction. The pith is loose parenchyma tissue and is mostly dead cells which may function in food storage when still alive.

In some woody stems the primary xylem may form a more or less continuous ring of tissue around the pith with no evident pith rays. In others, the primary xylem may occur as a ring of bundles surrounding the pith and separated from each

other by pith rays.

Descriptions of the Individual Stems as Seen in Cross-Section

Silver Poplar (*Populus alba*, Plate I)

Cork

Cork tissue of rather uniform, rectangular cells with those toward outer edge somewhat flattened, arranged in radial rows; not readily stained, some appearing almost impenetrable by the stain. Protoplasts evident in inner cells; outer cells sloughing off.

Cork cambium

Tissue apparently two cells in thickness, flattened between cork and cortex, stained deeply, showing protoplasts.

Cortex

Tissue of irregular parenchyma with darkly stained protoplasts and nuclei often evident; apparently two types of crystals, rhombohedral and rosette; the former more abundant in cells near pericycle.

Cortical fibers and stone cells

Bast fibers and stone cells in separate aggregates which tend to alternate throughout outer cortex. Stone cells large, with evident concentric stratifications. Bast fibers small, angular, with thick cell walls and small cavities.

Pericycle

Bast fibers and stone cells in alternate groups forming a narrow ring broken only by cortical ends of the rays. Bast fibers and stone cells like those of cortical bundles.

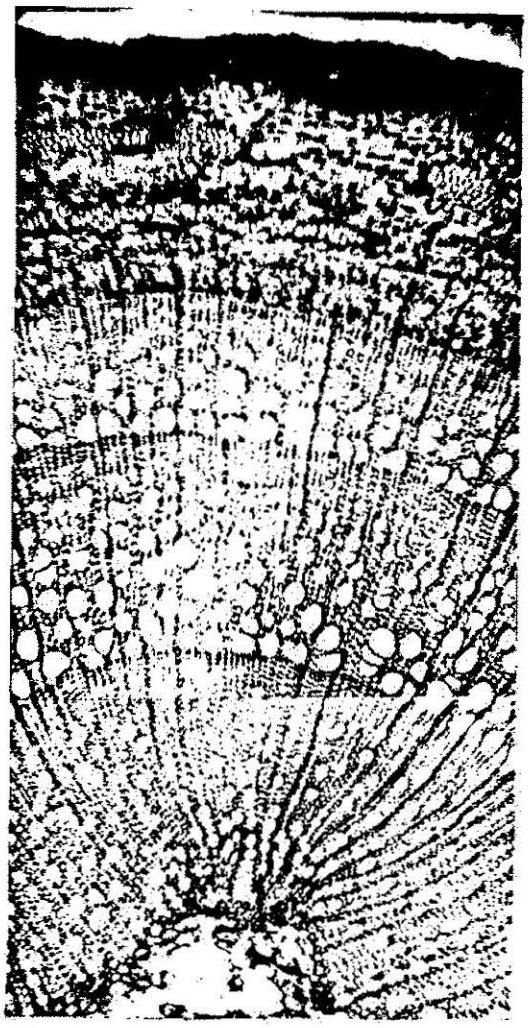

Plate I. Cross-Section of Silver Poplar (*Populus alba*) x 70

Phloem

Sieve tubes scattered; those of secondary phloem larger and more definite in outline than those of primary phloem. Companion cells smaller than sieve tubes; protoplasmic contents staining deeply. Groundwork of tissue, parenchyma.

Cortical ends of rays

Ends of the uniseriate vascular rays extending through phloem and pericycle. Cells larger and more box-like than those of phloem, staining more deeply.

Cambium

Several tangentially arranged rows of small deeply stained cells pressed closely together.

Xylem

Diffuse porous wood tissue; many vessels; wood fibers medium thick-walled; tracheids not distinct from fibers in cross section. Wood parenchyma present in uniseriate rays, among the last formed cells of annual ring, also around vessels of late spring growth. Cells of rays, long, narrow and pitted. Annual rings distinct with few or no vessels in summer wood; largest vessels in first spring growth.

Primary xylem

Tissue of the ring type in origin but some division into bundles; no pith rays. Each bundle consisting of radial series of small vessels surrounded by small thick walled parenchyma. Small bundles of sclerenchyma fibers occasionally found near pith.

Pith

Large parenchyma cells loosely held together. Living cells forming a sheath around tissue and extending into central portion, staining deeply; containing rosette crystals and starch.

Arizona Ash (Fraxinus velutina, Plate II)

Epidermis

Some epidermal cells and long slender hairs where tissue not ruptured.

Cork

Five or six rows of thin-walled, box-like cells; inner row of cells containing nuclei.

Cork cambium

Not distinct, just a region.

Cortex

Parenchyma cells, elongated tangentially, round on ends and flattened radially.

Pericycle

Alternating groups of sclerenchyma fibers and stone cells; fifty to sixty fibers in a group, five to ten stone cells, individual cells about five times as large as fibers. Stone cells with radiating markings; fibers thick-walled with small openings.

Phloem

Primary phloem rather crushed with scattering of parenchyma. Sieve tubes probably not active in this region. Uniseriate rays widening and spreading below pericycle.

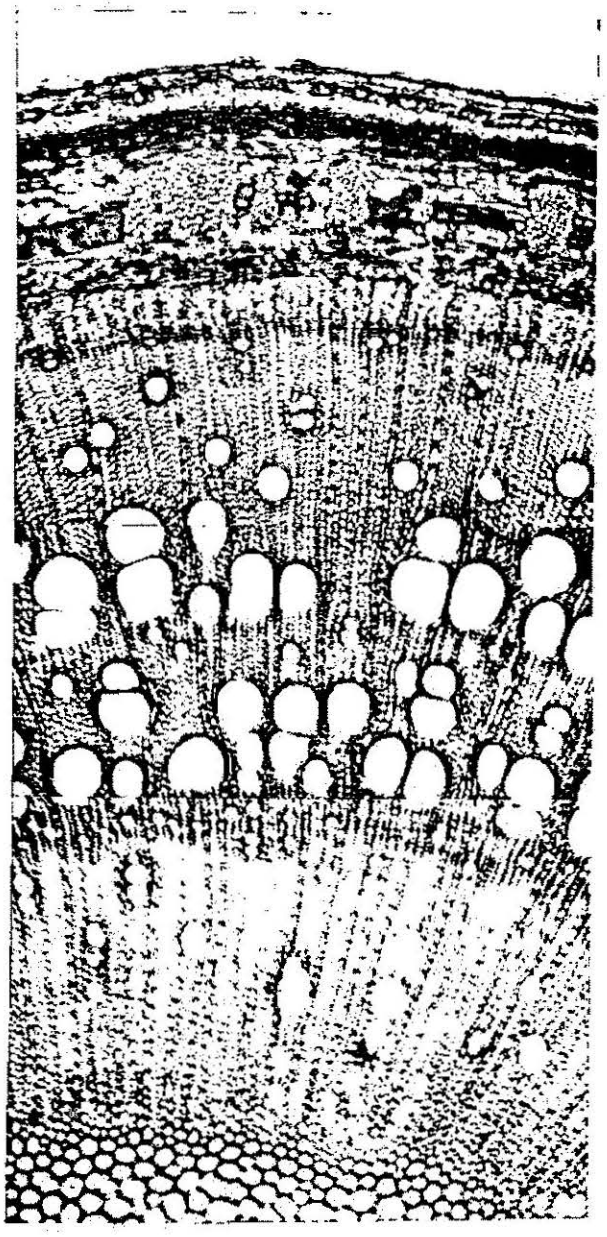

Plate II. Cross-Section of Arizona Ash
(*Fraxinus velutina*) x 70

In secondary phloem sieve tubes and companion cells. Sieve tubes irregularly thick-walled; openings variously shaped. Companion cells small, with evident nuclei; lying close to sieve tubes, especially those next to rays. Only other elements of tissue, broadened rays.

Cambium

Not definite as to cells; region 2 or 3 cells in thickness; cells crushed together and having dense protoplasmic contents.

Xylem

Wood fibers small, angular and compact with fairly heavy walls. Vessels large and grouped together in early spring wood; those of late spring and summer wood small, solitary or in radial groups of 2 or 3. Tracheids present, confined to the immediate vicinity of the vessels. Wood parenchyma paratracheal. Rays uniseriate and biseriate, numerous; rather large elongated cells with conspicuous nuclei. Scattered vessels of the inner region appearing to be infiltrated with a dark amorphous material. Some of the vessels with small globules of an unknown material protruding from the cell wall into the lumen of the vessel. Annual rings distinct; summer wood fibers and vessels smaller than spring elements.

Primary xylem

Not confined strictly to bundles but a cylinder of tissue only slightly grouped. Groups consisting of an inner portion of parenchyma and wood fibers with radially arranged

groups of small vessels; some of the vessels containing a dark resinous or perhaps tannin material.

Pith

Typical parenchyma; thin-walled, largely hexagonal and compact, with cellulose walls.

Silver Maple (Acer saccharinum, Plate III)

Cuticle and epidermis

Cuticle, of colorless and structureless substance following the contour of the epidermal cells and extending into the interstices between them. Epidermis, one layer of irregular cells.

Cork

Highly suberized cells not penetrated by the stains. Cells originally in radial rows; rounded to rectangular in shape tending to be longer than wide.

Cortex

Outer tissue collenchymatic. Parenchyma rounded; crystals in cells and intercellular spaces. Small scattered bundles of fibers, single fibers, and some stone cells.

Pericycle

Almost continuous band of fibrous tissue divided into long, irregular, compact bundles by parenchyma of rays. Stone cells occurring singly at infrequent intervals throughout tissue.

Phloem

Heterogeneous tissue; sieve tubes, parenchyma, solitary and grouped fibers and stone cells, and small nucleated com-

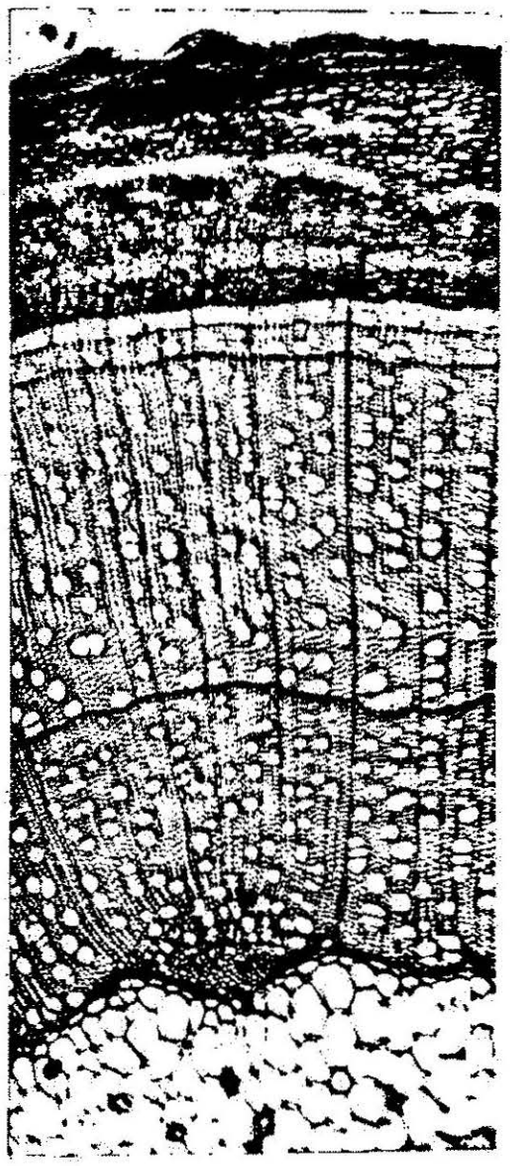

Plate III. Cross-Section of Silver Maple (*Acer saccharinum*) x 70

panion cells.

Secondary pericycle

Irregular, narrow band of tissue broken through by rays. Fiber cells with very small cavities.

Secondary phloem

Narrower and more dense than the primary phloem with fewer sieve tubes and fibers.

Cambium

Very narrow undifferentiated region.

Xylem

Wood diffuse porous; vessels scattered evenly throughout tissue, singly and in radial rows. Thick-walled wood fibers closely associated with vessels, those of rest of tissue mostly larger with thinner walls. Rays uniseriate, cells small and narrow, widening considerably in the phloem regions. Wood parenchyma paratracheal and metatracheal. Growth rings fairly distinct, delineated by a narrow dark line of dense fibrous tissue.

Primary xylem

Tissue in a ring of dome-shaped bundles. Base of the bundles of small thick-walled parenchyma. Vessels in groups of 4 or 5 increasing in size from the base; some of vessels apparently containing resinous substances. Rays and wood fibers between the vessel groups.

Pith

A double row of rather rounded deeply stained parenchyma ensheathing the pith. Remainder of tissue, large, loose,

angular cells some with crystals, some storage cells.

Sycamore (Platanus acerifolia, Plate IV)

Cuticle and epidermis

Remnants of a very thin cuticle; epidermal cells short and blunt.

Cork

Long rectangular to rounded cells in fairly even radial rows; cell walls thin; protoplasmic contents in most cells.

Cork cambium stained very deeply; can only be distinguished as a region.

Cortex

Irregularly arranged, variously shaped cells closely packed together. Protoplasmic contents and nuclei stained deeply. Rhombohedral crystals in some of the cells. Scattering of stone cells.

Pericycle

Compact, dome-shaped bundles of fibers alternating with stone cells and lying between the cortical ends of the rays.

Phloem

Crushed, dome-shaped aggregations of tissue lying between the cortical ends of the rays. Sieve tubes with thin, crumpled walls; companion cells small and rounded, few in number; scattering of parenchyma.

Cambium

Region merging imperceptibly with phloem on one side and xylem on the other.

Plate IV. Cross-Section of Sycamore
(Platanus acerifolia) x 70

Xylem

Wood diffuse porous; vessels occurring singly and in groups; larger and more numerous in first spring wood. Wood fibers thick-walled. Rays 3 - 14 seriate; largest ones originating in pith and dissecting the stele; smaller originating in primary xylem. Wood parenchyma paratracheal and metatracheal, paratracheal restricted to occasional cells, never forming a sheath; metatracheal abundant, scattered and in short irregular lines. Rings distinct, delineated by band of narrow, crowded wood fibers.

Primary xylem

Dome-shaped areas separated from each other by the wide rays. Base of each bundle small, thick-walled cells. Vessels grouped corresponding to vessels in rest of wood.

Pith

Oval and round cells with fairly thick walls. Many of the cells densely protoplasmic, probably storage cells.

Box Elder (Acer negundo, Plate V)

Cuticle and epidermis

Rather thick cuticle; epidermal cells small and pointed, the cuticle fitting into the interstices between the points.

Cork

Nothing properly suberized as yet; 2 or 3 rows of cells beginning to have appearance of cork cells.

Cortex

Fairly homogeneous, compact tissue of slender, flattened cells with rounded ends. Scattering of large rosette crys-

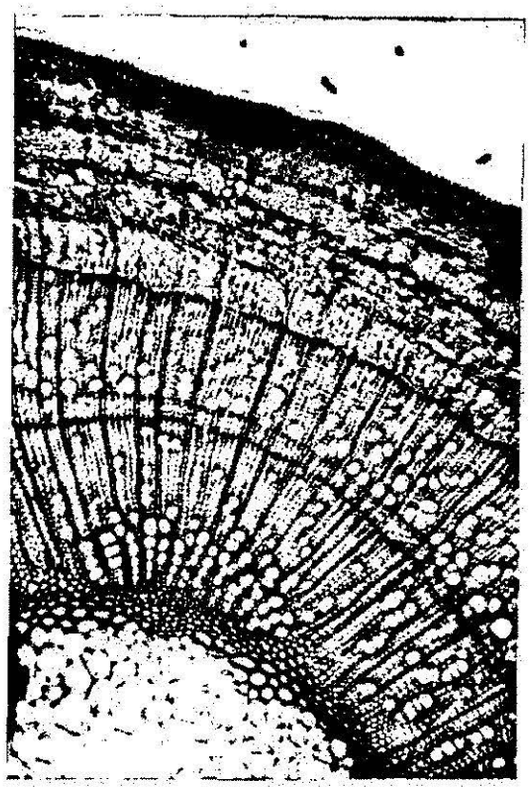

Plate V. Cross-Section of Box Elder
(Acer negundo) x 70

tals in intercellular spaces; other crystals in cells close to pericycle.

Pericycle

Alternating bundles of fibers and stone cells in an irregular band around the stem. Fibers small; stone cells varying in size and shape, markings evident.

Phloem

In primary phloem small parenchyma cells, sieve tubes, some crystals and stone cells. Sieve tubes conspicuous with irregularly thickened walls and large openings. Rays spreading from one cell wide to several in this tissue. Secondary phloem, largely parenchyma. Sieve tubes in formation, still containing photoplasts. Rays uniseriate.

Cambium

Several rows of very small cells tightly compressed.

Xylem

Vessels rather diffuse, in groups; larger and more plentiful in early spring wood. Wood fibers immediately surrounding vessels with thick walls; others with medium thick walls. Rays 1 to 3 seriate of short slender cells. Wood parenchyma, metatracheal. Annual ring quite evident; delineated by small very thick-walled summer fibers.

Primary xylem

Ring of tissue with scattered large bundles, consisting of chains of vessels grading from small to large surrounded by small thick-walled parenchyma.

Pith

Complete ring of functional parenchyma tissue encircling pith. Inner pith, large, loose, non-functional parenchyma with scattering of rosette crystals.

Blue Elder (Sambucus glauca, Plate VI)

Cuticle and epidermis

Tissues ruptured and broken away in most places by formation of cork beneath. Cuticle thin; epidermal cells densely protoplasmic.

Cork

Cork two or three cells in thickness; cells almost square, tending to be longer in radial direction. Tissue bunching and thick at lenticels.

Cortex

Band of small, round, irregularly thick-walled collenchyma comprising outer part of tissue. Cortical parenchyma large, elongated, nuclei and cell contents staining deeply.

Pericycle

Long, narrow bundles of large fibers encircling stem.

Phloem

Primary phloem tissue consisting of round thick-walled parenchyma, irregularly-shaped sieve tubes, and a scattering of darkly stained cells containing tannin or some such substance. Rays merging with parenchyma of tissue. Secondary phloem, same elements as primary, smaller and more crowded, parenchyma confined to rays. Secondary bundles of fibers occurring between primary and secondary phloem.

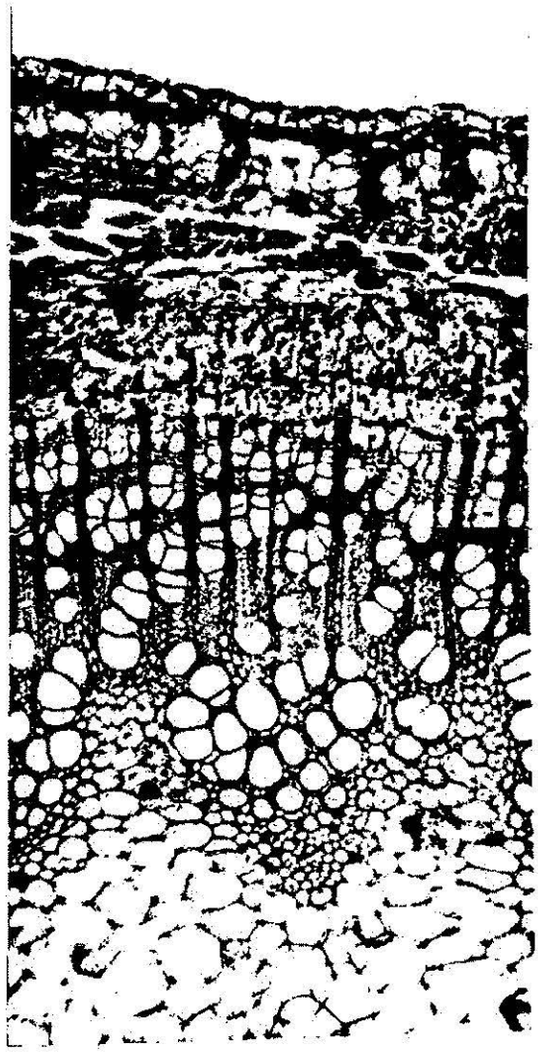

Plate VI. Cross-Section of Blue Elder (Sambucus glauca) x 70

Cambium

Not distinct.

Xylem

Whole tissue having a netted appearance due to grouping of vessels of varying sizes. Groups tending to be in tangential rows. Wood fibers very thick-walled. Rays 1 to 6 seriate. Wood parenchyma metatracheal.

Primary xylem

Large, dome-shaped bundles of tissue extending into pith. Base of bundles small thick-walled parenchyma. Vessels larger than in secondary xylem. Rays between bundles quite wide.

Pith

Large, loose parenchyma; scattering of crystals and storage cells.

Weeping Willow (Salix babylonica, Plate VII)

Cuticle and epidermis

Thin cuticle and small epidermal cells in some places.

Cork

Two to several rows of thin-walled, flattened cells, inner cells heavily suberized.

Cortex

Oval and rounded parenchyma; nuclei and protoplasts staining deeply. Intercellular spaces and rosette crystals scattered throughout.

Pericycle

Scattered bundles of fibers; not a continuous tissue.

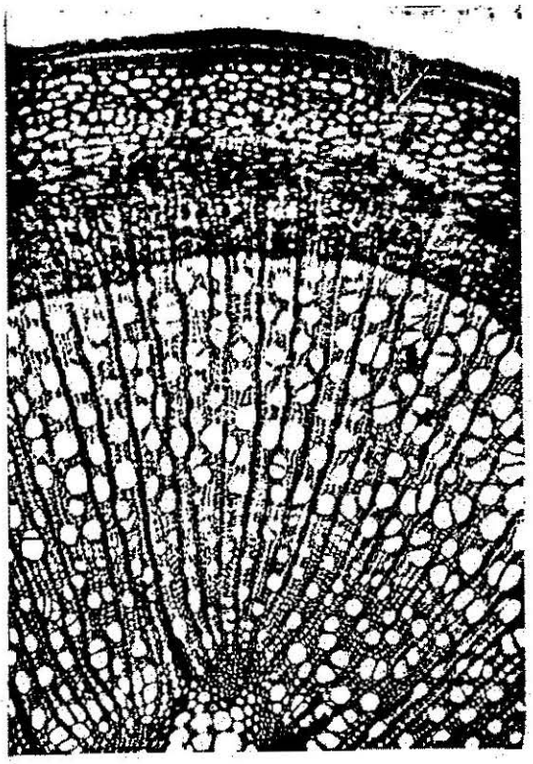

Plate VII. Cross-Section of Weeping Willow (*Salix babylonica*) x 70

Phloem

Primary and secondary tissue consisting of parenchyma, sieve tubes, bundles of fibers, some crystals and dark cells containing a material like tannin, and a few small nucleated companion cells. Uniseriate rays with large cells in this region.

Cambium

A very narrow region, cells not distinct.

Xylem

Diffuse porous wood; vessels grouped or single, abundant. Wood fibers with large openings, thin walls. Wood parenchyma mostly confined to uniseriate, branching rays. Growth ring indistinct; little difference between spring and summer woods.

Primary xylem

Ring of tissue irregularly sheathing pith; nearly continuous; some division into bundles. Bundles consisting of basal, small parenchyma and radial chains of vessels; parenchyma extending between the vessel groups as rays.

Pith

Sheath of large parenchyma around pith; cells staining more deeply than other pith cells, granular appearance. Inner cylinder of pith typical large, loose cells.

Black Locust (Robinia pseudoacacia, Plate VIII)

Cork

Crushed suberized tissue several layers thick. Separated from cortex by layer of loose, easily ruptured phelloderm.

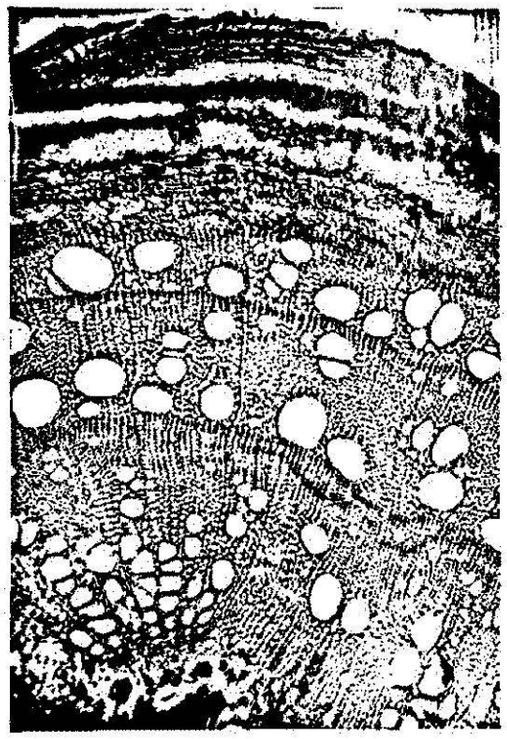

Plate VIII. Cross-Section of Black Locust (Robinia pseudoacacia) x 70

Cortex

Irregular band of tissue of rounded parenchyma; outer cells slightly suberized. Tissue somewhat broken by layers of phelloderm. Some rhombohedral crystals.

Pericycle

Large bundles of fibers, alternating with small groups of stone cells. Fibers very compact; stone cells with stratifications. Many crystals around edge of tissue and with stone cells.

Phloem

Tissue very compressed and irregular. Some sieve tubes with protoplasmic contents, some open cells, others collapsed leaving nothing but an irregular band of structureless substance. Parenchyma confined to uniseriate rays. In outer region of tissue scattering of cells containing dark amorphous substance. Many bundles of fibers and crystals throughout tissue.

Cambium

Tissue 2 or 3 cells in thickness; cells very small, compressed, with stained nuclei.

Xylem

Vessels occurring in clusters, those of spring wood rather large; tyloses in some of the vessels. Wood fibers, small, fairly thick walls. Rays uniseriate, thin, cells small. Wood parenchyma abundant, terminal and paratracheal, crystals in nearly all cells. Rings distinct; transition from spring to summer wood rather abrupt; summer wood vessels

small, scarce.

Primary xylem

Ring of bundles consisting of basal portion of slightly lignified cells extending between chain-groups of large vessels.

Pith

Outer pith containing many crystals in pith parenchyma and clusters of large cells filled with a dark material. Inner pith, large, spongy cells, easily torn apart.

Virginia Creeper (Ampelopsis sp., Plate IX)

Cork

Tissue thick and uneven; cells thin-walled, rectangular.

Cortex

Typical cortical parenchyma; staining deeply; many of cells almost obliterated by dark substance within. Frequent tubular cells in tissue with remains of secretion in them. Joining cortex are wide funnel-shaped ends of rays.

Pericycle

Small compact bundles of fibers lying between ends of rays.

Phloem

Tissue wide radially, lying between the rays. Great deal of parenchyma, many containing dark substance, and sieve tubes. Bundles of fibers variously arranged throughout tissue. Rays funnel-shaped, narrowing to cambium; basically parenchyma; containing some tubular cells as in cortex, and large cells containing bundles of raphide crystals.

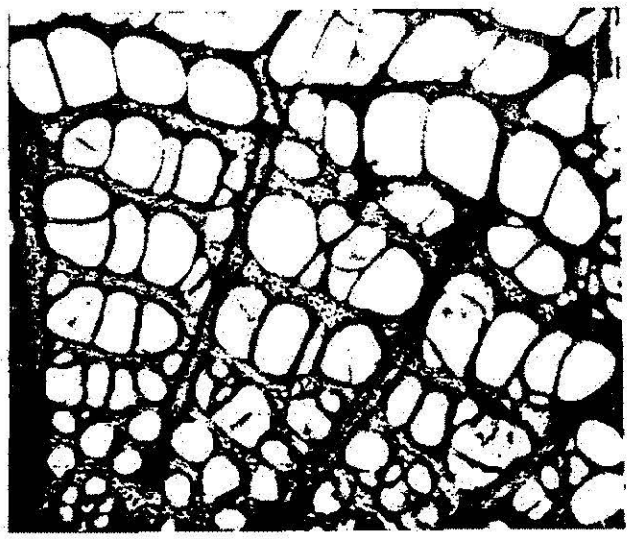

Plate IX. Cross-Section of Xylem of Virginia Creeper (*Ampelopsis* sp.) showing Tyloses. x 70

Xylem

Tissue consisting of large vessels, grouped together by very thick-walled wood fibers into tangential rows. Proportion of wood fiber increasing toward center and vessels smaller. Vessels of inner half of tissue containing tyloses, many of vessels completely filled. Wood parenchyma paratracheal, but not abundant. Rays, many seriate; cells long, slender with flattened ends, evident nuclei and pitted walls.

Primary xylem

Stele dissected by the rays into radial bands of tissues. At base of each band wood fibers and ray parenchyma merging into thick walled cells with some characteristics of both; many containing a dark substance. Inconspicuous bundles of small vessels in this tissue, original vessels of stem.

Pith

Large, round pith parenchyma and tubular cells. Many of parenchyma containing dark substance giving tissue a checkerboard appearance.

CHAPTER IV

SUMMARY AND CONCLUSIONS

Stem for General Use

Study and comparison of the various stems described, Silver Poplar, Arizona Ash, Silver Maple, Sycamore, Box Elder, Blue Elder, Weeping Willow, Locust, and Virginia Creeper has been made. Consideration of their desirable qualities for class use indicates the following:

The Silver Poplar, Arizona Ash, Silver Maple, Sycamore, and Box Elder, have the most "typical" kinds of woody tissues. All of these stems can be cut and stained quite readily. Their tissues are arranged in a similar manner, the principal differences being in amounts of tissues and cell-types.

The Arizona Ash appears to be the best of those studied for general class work, for the following reasons:

1. The material cuts and stains readily.
2. The stain clearly differentiating the tissues.
3. The content, size and structure of the tissues are such that all cells are penetrated by the stains and their character easily seen.
4. The tissues and individual cells can be readily distinguished under the low power of the microscope.
5. The arrangement of the tissues is simple and diagrammatic with each tissue clearly delimited, so as to be readily recognizable by the student with the aid of a brief description.

Stems for Special Uses

Several stems showed certain tissues which are well adapted to demonstration and projection to illustrate cell-types.

The Locust stem shows a good growth of cork, a peculiar phloem tissue, and an abundant and interesting arrangement of wood parenchyma and crystals. It does not take stains readily and has a tendency to tear apart in cutting.

The Blue Elder shows a dissected stele and collenchyma tissue in the cortex. It does not cut readily, as the outer bark tissues tend to break away from the vascular tissues.

The Virginia Creeper stem is interesting as a woody dicotyledonous vine-type. The very wide expansion of the upper ends of the rays, the bundles of crystals and the tyloses in the numerous vessels, would be desirable for demonstrations in botany classes.

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